A P P E N D I X

Marked-Up Version of Changes

IN THE SPECIFICATION:

The specification is amended as follows:

Page 26, line 15 to Page 27, line 3, are amended as follows:

"Chromosomal DNA was prepared from bacteria grown overnight at 37°C in a volume of 30 mL of Luria broth. After harvesting by centrifugation, cells were washed and resuspended in 10 mL of [50mMTris-HCl] 50 mM Tris-HCl pH 8.0. EDTA was added and the mixture incubated for 20min. Then lysozyme was added and incubation continued for a further 10min. Proteinase K, SDS, and ribonuclease were then added and the mixture incubated for up to 2hr for lysis to occur. All incubations were at 37°C. The mixture was then heated to 65°C and extracted once with 8mL of phenol at the same temperature. The mixture was extracted once with 50mL of phenol/chloroform/iso-amyl alcohol at 4°C. Residual phenol was removed by two ether extractions. DNA was precipitated with 2 vols. of ethanol at 4°C, spooled and washed in 70% ethanol, resuspended in 1-2mL of TE and dialysed. Plasmid and cosmid DNA was prepared by a modification of the Birnboim and Doly method (Birnhoim, H. C. And Doly, J. (1979) A rapid alkaline extraction procedure for screening recombinant plasmid DNA *Nucl. Acid Res.* 7:1513-1523. The volume of culture was 10nmL and the lysate was extracted with phenol/chloroform/iso-amyl alcohol before precipitation with isopropanol. Plasmid DNA to be used as vector was isolated on a continuous caesium chloride gradient following alkaline lysis of cells grown in 1L of culture."

Page 27, lines 22-32, are amended as follows:

"Colonies were screened for the presence of the O111 antigen by immunblotting. Colonies were grown overnight, up to 100 per plate then transferred to nitrocellulose discs and lysed with 0.5N HCl. Polyoxyethylene(20)-sorbitan monolaurate (TWEEN [Tween] 20) was added to TBS at 0.05% final concentration for blocking, incubating and washing steps. Primary antibody was *E. coli* O group 111 antiserum, diluted 1:800. The secondary antibody was goat anti-rabbit IgG labelled with horseradish peroxidase diluted 1:5000. The staining substrate was 4-chloro-1-naptol. Slide agglutination was performed according to the standard procedure."

Page 34, lines 6-25, are amended as follows:

"*E. coli* O157 O antigen gene cluster was amplified by using long PCR (Cheng et al. 1994, "Effective amplification of long targets from cloned inserts and human and genomic DNA" P.N.A.S. USA 91: 5695-569) with one primer (primer #412: att ggt agc tgt aag cca agg gcg gta gcg t (SEQ ID NO:5)) based on the JumpStart sequence usually found in the promoter region of O antigen gene clusters (Hobbs, *et al.* 1994 "The JumpStart sequence: a 39 bp element common to several polysaccharide gene clusted" Mol. Microbiol. 12: 855-856), and another primer #482 (cac tgc cat acc gac gac gcc gat ctg ttg ctt gg (SEQ ID NO:6)) based on the *gnd* gene usually found downstream of the O antigen gene cluster. Long PCR was carried out using the Expand Long Template PCR System from Boehringer Mannheim (Castle Hill NSW Australia), and products, 14 kb in length, from several reactions were combined and purified using the Promega Wizard PCR preps DNA purification System (Madison WI USA). The PCR product was then extracted with phenol

and twice with ether, precipitated with 70% ethanol, and resuspended in 40 µL of water."

Pages 94-103, renumber as pages 61-69, respectively.

IN THE CLAIMS:

The claims are amended as follows:

Claim 85. (Amended) A method of testing a sample for the presence of *E. coli* expressing the bacterial polysaccharide O-antigen serotype 0111, the method comprising the steps of:

(a) providing genomic DNA of a sample to be tested;

(b) providing at least one oligonucleotide molecule, wherein said oligonucleotide molecule is at least about 10 nucleotides in length, and [specifically] hybridizes using high stringent wash conditions to a nucleic acid sequence selected from the group consisting of:

wbdH (nucleotide positions 739 to 1932 of SEQ ID NO: 1);

wzx (nucleotide positions 8646 to 9911 of SEQ ID NO: 1);

wzy (nucleotide positions 9901 to 10953 of SEQ ID NO: 1); and

wbdM (nucleotide positions 11821 to 12945 of SEQ ID NO: 1),

wherein said high stringent wash conditions consists of 3 x 5 min washes in 2 x SSC and 0.1% SDS at room temperature, a 1 hr wash in 1 x SSC and 0.1% SDS at 58°C and 15 min wash in 0.1 x SSC and 0.1% SDS at 58°C;

(c) contacting said genomic DNA with said at least one oligonucleotide molecule [under conditions suitable] to permit said oligonucleotide molecule to [specifically] hybridize under said high strigent wash conditions to said nucleic acid sequence when present in said genomic DNA; and

(d) detecting any [specifically] hybridized oligonucleotide molecules, wherein detection of said hybridized oligonucleotide molecules indicates the presence of said *E. coli* in said sample.

Claim 88. (Amended) The method as claimed in Claim 85, wherein said at least one oligonucleotide molecule is selected from the group consisting of [the oligonucleotides listed in tables 5 and 5A herein] positions 739-757 of SEQ ID NO:1, positions 925-942 of SEQ ID NO:1, positions 1165-1182 of SEQ ID NO:1, positions 8646-8663 of SEQ ID NO:1, positions 8906-8923 of SEQ ID NO:1, positions 9150-9167 of SEQ ID NO:1, positions 9976-9996 of SEQ ID NO:1, positions 10113-10130 of SEQ ID NO:1, positions 11821-11844 of SEQ ID NO:1, positions 12042-10259 of SEQ ID NO:1, positions 12258-12275 of SEQ ID NO:1, positions 1941-1924 of SEQ ID NO:1, positions 1731-1714 of SEQ ID NO:1, positions 1347-1330 of SEQ ID NO:1, positions 9908-9891 of SEQ ID NO:1, positions 9468-9451 of SEQ ID NO:1, positions 9754-9737 of SEQ ID NO:1, positions 10827-10807 of SEQ ID NO:1, positions 10484-10467 of SEQ ID NO:1, positions 12945-12924 of SEQ ID NO:1, positions 12447-12430 of SEQ ID NO:1 and positions 12698-12681 of SEQ ID NO:1.

Claim 89. (Amended) A method of testing a sample for the presence of *E. coli* expressing the bacterial polysaccharide O-antigen serotype 0157, the method comprising the steps of:

(a) providing genomic DNA of a sample to be tested;

(b) providing at least one oligonucleotide molecule, wherein said oligonucleotide molecule is at least about 10 nucleotides in length and [specifically] hybridizes using high stringent wash conditions to a nucleic acid sequence selected from the group consisting of:

wbdN (nucleotide position 79 to 861 of SEQ ID NO: 2);

wbdO (nucleotide positions 2011 to 2757 of SEQ ID NO: 2);

wbdP (nucleotide positions 5365 to 6471 of SEQ ID NO: 2);

wbdR (nucleotide positions 13156 to 13821 of SEQ ID NO: 2);

wzx (nucleotide positions 2744 to 3109 of SEQ ID NO: 2); and

wzy (nucleotide positions 858 to 2042 of SEQ ID NO: 2),

wherein said high stringent wash conditions consists of 3 x 5 min washes in 2 x SSC and 0.1% SDS at room temperature, a 1 hr wash in 1 x SSC and 0.1% SDS at 58°C and 15 min wash in 0.1 x SSC and 0.1% SDS at 58°C;

(c) contacting said genomic DNA with said at least one oligonucleotide molecule [under conditions suitable] to permit said oligonucleotide molecule

to [specifically] hybridize under said high strigent wash conditions to said nucleic acid sequence when present in said genomic DNA; and

(d) detecting any specifically hybridized oligonucleotide molecules, wherein detection of said hybridized oligonucleotide molecules indicates the presence of said *E. coli* in said sample.

Claim 92. (Amended) The method as claimed in Claim 89, wherein said at least one oligonucleotide molecule is selected from the group consisting of [the oligonucleotides listed in tables 6 and 6a herein] positions 79-96 of SEQ ID NO:2, positions 184-201 of SEQ ID NO:2, positions 310-327 of SEQ ID NO:2, positions 858-875 of SEQ ID NO:2, positions 1053-1070 of SEQ ID NO:2, positions 1278-1295 of SEQ ID NO:2, positions 2011-2028 of SEQ ID NO:2, positions 2110-2127 of SEQ ID NO:2, positions 2305-2322 of SEQ ID NO:2, positions 2744-2761 of SEQ ID NO:2, positions 2942-2959 of SEQ ID NO:2, positions 5440-5457 of SEQ ID NO:2, positions 5707-5724 of SEQ ID NO:2, positions 13261-13278 of SEQ ID NO:2, positions 13384-13401 of SEQ ID NO:2, positions 861-844 of SEQ ID NO:2, positions 531-514 of SEQ ID NO:2, positions 768-751 of SEQ ID NO:2, positions 2042-2025 of SEQ ID NO:2, positions 1619-1602 of SEQ ID NO:2, positions 1913-1896 of SEQ ID NO:2, positions 2757-2740 of SEQ ID NO:2, positions 2493-2476 of SEQ ID NO:2, positions 2682-2665 of SEQ ID NO:2, positions 6471-6454 of SEQ ID NO:2, positions 5973-5956 of SEQ ID NO:2, positions 6231-6214 of SEQ ID NO:2, positions 13629-13612 of SEQ ID NO:2 and positions 13731-13714 of SEQ ID NO:2.

Claim 93. (Amended) A method of testing a sample for the presence of *S. enterica* expressing the bacterial polysaccharide O-antigen serotype C2, the method comprising the steps of:

(a) providing genomic DNA of a sample to be tested;

(b) providing at least one oligonucleotide molecule, wherein said oligonucleotide molecule is at least about 10 nucleotides in length and [specifically] hybridizes using high stringent wash conditions to a nucleic acid sequence selected from the group consisting of:

[*weaR*] *wbaR* (nucleotide positions at 2352 to 3314 of SEQ ID NO: 3);

wbaL (nucleotide positions 3361 to 3875 of SEQ ID NO: 3);

wbaQ (nucleotide positions 3977 to 5020 of SEQ ID NO: 3);

wbaW (nucleotide positions 6313 to 7323 of SEQ ID NO: 3);

wbaZ (nucleotide positions 7310 to 8467 of SEQ ID NO: 3);

wzx (nucleotide positions 1019 to 2359 of SEQ ID NO: 3); and

wzy (nucleotide positions [5144] 5114 to 6313 of SEQ ID NO: 3),

wherein said high stringent wash conditions consists of 3 x 5 min washes in 2 x SSC and 0.1% SDS at room temperature, a 1 hr wash in 1 x SSC and 0.1% SDS at 58°C and 15 min wash in 0.1 x SSC and 0.1% SDS at 58°C;

(c) contacting said genomic DNA with said at least one oligonucleotide molecule [under conditions suitable] to permit said oligonucleotide molecule to [specifically] hybridize under said high strigent wash conditions to said nucleic acid sequence when present in said genomic DNA; and

(d) detecting any specifically hybridized oligonucleotide molecules, wherein detection of said hybridized oligonucleotide molecules indicates the presence of said *S. enteria* in said sample.

Claim 96. (Amended) The method as claimed in Claim 93, wherein said at least one oligonucleotide molecule is selected from the group consisting of [the oligonucleotides listed in table 7 herein] positions 1019-1036 of SEQ ID NO:3, positions 1708-1725 of SEQ ID NO:3, positions 1938-1955 of SEQ ID NO:3, positions 2352-2369 of SEQ ID NO:3, positions 2601-2618 of SEQ ID NO:3, positions 2910-2927 of SEQ ID NO:3, positions 3361-3378 of SEQ ID NO:3, positions 3578-3595 of SEQ ID NO:3, positions 3977-3994 of SEQ ID NO:3, positions 4167-4184 of SEQ ID NO:3, positions 4603-4620 of SEQ ID NO:3, positions 5114-5131 of SEQ ID NO:3, positions 5664-5681 of SEQ ID NO:3, positions 5907-5924 of SEQ ID NO:3, positions 6313-6330 of SEQ ID NO:3, positions 6697-6714 of SEQ ID NO:3, positions 6905-6922 of SEQ ID NO:3, positions 7310-7327 of SEQ ID NO:3, positions 7530-7547 of SEQ ID NO:3, positions 8007-8024 of SEQ ID NO:3, positions 1414-1397 of SEQ ID NO:3, positions 2170-2153 of SEQ ID NO:3, positions 2356-2339 of SEQ ID NO:3, positions 2759-2742 of SEQ ID NO:3, positions 3047-3030 of SEQ ID NO:3, positions 3311-3294 of SEQ ID NO:3, positions 3759-3742 of SEQ ID NO:3, positions 4378-4361 of SEQ ID

NO:3, positions 4774-4757 of SEQ ID NO:3, positions 5017-5000 of SEQ ID NO:3, positions 5515-5498 of SEQ ID NO:3, positions 6112-6095 of SEQ ID NO:3, positions 6310-6293 of SEQ ID NO:3, positions 6805-6788 of SEQ ID NO:3, positions 7068-7051 of SEQ ID NO:3, positions 7320-7303 of SEQ ID NO:3, positions 7775-7758 of SEQ ID NO:3, positions 7907-7890 of SEQ ID NO:3 and positions 8464-8447 of SEQ ID NO:3.

Claim 97. (Amended) A method of testing a sample for the presence of *S. enterica* expressing the bacterial polysaccharide O-antigen serotype B, the method comprising the steps:

(a) providing genomic DNA of a sample to be tested;

(b) providing at least one oligonucleotide molecule, wherein said oligonucleotide molecule is at least about 10 nucleotides in length and [specifically] hybridizes using high stringent wash conditions to a nucleic acid sequence selected from the group consisting of:

wzx (nucleotide positions 12762 to 14054 of SEQ ID NO: 4); and

wbaV (nucleotide positions 14059 to 15060 of SEQ ID NO: 4),

wherein said high stringent wash conditions consists of 3 x 5 min washes in 2 x SSC and 0.1% SDS at room temperature, a 1 hr wash in 1 x SSC and 0.1% SDS at 58°C and 15 min wash in 0.1 x SSC and 0.1% SDS at 58°C;

(c) contacting said genomic DNA with said at least one oligonucleotide molecule [under conditions suitable] to permit said oligonucleotide molecule to [specifically] hybridize under said high

strigent wash conditions to said nucleic acid sequence when present in said genomic DNA; and

(d) detecting any specifically hybridized oligonucleotide molecules, wherein detection of said hybridized oligonucleotide molecules indicates the presence of said *S. enteria* in said samples.

Claim 100. (Amended) The method as claimed in Claim 97, wherein said at least one oligonucleotide molecule is selected from the group consisting of [the oligonucleotides listed in table 8 herein] positions 12762-12779 of SEQ ID NO:4, positions 12993-13010 of SEQ ID NO:4, positions 13635-13652 of SEQ ID NO:4, positions 14059-14076 of SEQ ID NO:4, positions 14688-14705 of SEQ ID NO:4, positions 13150-13133 of SEQ ID NO:4, positions 13417-13400 of SEQ ID NO:4, positions 14051-14034 of SEQ ID NO:4, positions 14421-14404 of SEQ ID NO:4, and positions 15057-15040 of SEQ ID NO:4.

IN THE SEQUENCE LISTING:

Pages 61-93 (Sequence Listing), delete in their entirety.